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A Study Of Liquid Fertilizers

A STUDY OF LIQUID FERTILIZERS

BY

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I. Introduction

In the growing of many floricultural and other green house plants, it is often inadvisable or impossible to incorporate in the soil at the beginning sufficient available food to maintain the plants in the best condition throughout the season and secure a maximum production. Consequently feeding at varying intervals is necessary. For such feeding, liquid fertilizers are easily applicable and have been quite widely used.

One type of liquid fertilizer is a solution of various "commercial fertilizers" containing the proper proportion of the necessary elementary constituents. This type has many advantages. It can be made of a desired composition. It is readily obtainable and can be quickly, easily and cheaply prepared and applied. However, since the constituents are in a readily available form, there is a great tendency toward over-feeding. The untoward results which have accompanied the over feeding in many cases have brought about a hesitancy in the use of commercial fertilizers.

Another type of liquid fertilizer is an aqueous extract of the solid excrement of cows, horses, sheep and other animals. This type has an advantage over a "commercial" fertilizer in that certain organic substances are present whose decomposition

produces on the soil conditions and plant growth a beneficial effect in addition to the simple food elements which are supplied. However in large cities, it is already becoming a serious problem to secure a good grade of manure at all times. Moreover, in any case the composition of the manure is quite variable and consequently that of the extract is also. The preparation of the extract involves more labor and inconvenience than a solution of the commercial fertilizers, and the liquid is less easily pumped through a green house and applied to the soil.

Concerning the fertilizing content of liquid manure, very little has been reported in the Experiment Station Record. The published analytical work[1] is for the most part confined to urine and such liquid manure as would ordinarily drain from a stable. This is of a considerably different type from that which is used in floricultural growing. Consequently an analytical study of this latter type was first necessary to determine the fertilizing content. In order to place the study on as practical a basis as possible before beginning the investigation, an effort was made to find out the general methods of preparation and application of liquid manure which are used by the florists of the state and the results of their use. A blank request for information was sent to about forty different floricultural establishments. Only a few replies were received and the information received was very indefinite. Opinions varied as to the utility of liquid manure, and on the

whole the data obtained was very unsatisfactory and incomplete.

The following methods seemed the most representative:

A certain quantity of solid excrement is treated with water and the extract made as follows:

1) The mixture is stirred thoroughly, allowed to settle and the extract drawn off at once.

2) The mixture is heated

a) by a steam coil or

b) by passing steam through it and then drawing off the extract.

3) The mixture is allowed to ferment for a time before extraction.

4) The first extraction is made as 1) then the residue again made up with water and allowed to ferment and a second extraction made.

Heating would perhaps have a tendency to give a better solution or suspension of the material and would break down some of the more unstable compounds in the manure. However it would also promote loss of ammonia by volatilisation. In addition it would bring about at least a partial sterilization.

Fermentation affords an opportunity for decomposition of the organic matter, chiefly anerobically and would affect the availability after application. At the same time there would be opportunity for loss of ammonia by volatilization, loss of nitrogen by denitrification and perhaps the formation of some substance which might hinder plant growth.

II. Experimental

Laboratory Study of the Methods of Preparation of Liquid Manure

In the study of the methods of preparation of the extract, fresh cow manure, since it is used at this station and most generally elsewhere by florists who can obtain it, was subjected to a detailed investigation. The usual quantities employed are in the proportion of one half bushel of manure to fifty gallons of water or approximately 250 grams per 2300cc. water.

The manure used was obtained fresh, and as free from litter as possible, at the University dairy barns.

The following general method of procedure was carried out:

The fresh sample of manure was thoroughly mixed and portions dried at 100 degrees for moisture determination. After drying the sample was ground and analyzed.

Other fresh portions of about 250 grams each were weighed out into 2300cc. Jena flasks. About 1500-2000cc. of distilled water was then added and after shaking and mixing thoroughly the treatment recorded below was made.

Portions of the sample for the cold extractions V-E, VI-E, VII-E, VIII-E, IX-E, and IX-F (see table I - VI) were made up to volumes of 2300cc. and the extract filtered through cotton gauze and weighed. The weight in grams was recorded as

cc. volume. The residue was returned to the flask and again made up to volume with distilled water. They were stoppered and set away to incubate at room temperature, numbers V-E, VI-E, and VII-E for a period of two weeks, and number VIII-E for a period of seven weeks. Extraction was again made as before and the residue dried and analyzed.

Portions for the cold extraction IX-G and IX-H were made up to volume and allowed to ferment at room temperature in a stoppered flask for four weeks before an extraction was made.

After the 1500-2000cc. of water had been added to portions V-A, VI-A, and VII-A for hot extraction, they were then heated at the boiling point for about thirty minutes, cooled, made up to volume and extracted as before. Portion VIII-A was heated to the boiling point and maintained boiling gently for 30 minutes. VIII-C was heated to boiling and steam passed through for 30 minutes.

IX-A and B were heated only to the boiling point, about 30 minutes being consumed in the process.

IX-C was heated to the boiling point, then steam was passed through rapidly for 30 minutes.

After the above treatments the flasks were cooled, made up to volume and extractions made as usual.

Solids were determined in the extracts by evaporation of 100cc. portions on the water bath at 100°.

The fertilizing constituents determined were total nitrogen, phosphorus, and potassium. In all cases duplicates were run.

Nitrogen was determined in the 100cc. portions of the extract and 2 gram samples of the dry manure and residue by the official Kjeldahl method.

Phosphorus. After the addition of a few cc. of alcoholic calcium acetate solution to the solids they were evaporated to dryness and ignited, taken up in dilute nitric acid, filtered, and the phosphate precipitated as the phosphomolybdate, then dissolved in ammonia, precipitated with magnesia mixture, and determined as magnesium pyrophosphate by the official method.[2]

For the original sample and residue, 2 gram portions of the ground material were taken and sufficient calcium acetate solution added to moisten thoroughly before ignition, and the determination completed as above.

Potassium. 2 gram samples of the original manure and residue (and 100cc. portions of extract evaporated to dryness) were ignited at a low red heat, taken up in hot water, filtered, heated to boiling and barium hydroxide added until no further precipitate formed, filtered hot, the filtrate cooled and ammonium hydroxide and ammonium carbonate added to precipitate the excess of barium. The barium carbonate was filtered out and the filtrate evaporated to dryness, ignited to remove ammonium salts, taken up with a drop of hydrochloric acid and filtered to remove any carbonaceous matter. Chloroplatinic acid was then added and the potassium determined as potassium chlorplatinate, after evaporation almost to dryness and washing out the sodium salt with alcohol.

Free Ammonia. Several attempts were made to determine the loss of ammonia in heating and boiling the extracts, by drawing air through a stopper in the flasks over the liquid and into standard acid. The method gave no appreciable amounts of ammonia.

Potassium in the extract was in some instances determined by difference of the total in the residue from the total in the original material extracted.

Residue. The weight of the residue was determined by subtraction of the weight of the total solids of the extracts from the dry weight equivalent of the material extracted. In some cases the residue was removed from the filtering cloth as completely as possible and weighed after drying at 100°.

Discussion of the results of the various methods of extraction

The results of the analytical study of the methods of extraction are given in tables I - VI. A summation of the total percentages of the various constituents in the residue and extract usually slightly exceeds the 100% of the original manure. However there are several almost unavoidable sources of error. Some loss of ammonia must take place in the drying of the original manure. The determination of the residue by difference from the solids on evaporation of the organic extract is not entirely satisfactory and accurate but affords a means of checking the results to a certain degree.

Since the various samples of the original manure vary considerably in fertilizing content it is to be expected that the extracts will also. However the variation in the percentages of the total constituents extracted is proportionally as great as the variation between the different samples.

The extraction treatment A and B represents the heating of the aqueous mixture at the boiling point for about thirty minutes before filtration. Treatment C represents heating by passing steam through the mixture for about thirty minutes, treatment E and F the immediate cold extraction, G and H the cold extraction after several weeks, and E^1 the second cold extraction following E after a period of a few weeks fermentation.

Table 1

Analysis of Original Manure Used for Extraction

Sample		V	VI	VII	VIII	IX
Date of Collection		11/24 1914	12/4 1914	1/6 1915	3/31 1915	9/14 1915
% Moisture		81.6	79.35	8.36	84.9	82.4
% Dry Weight		18.4	20.65	16.40	15.1	17.6
% Dry Weight	(N	2.15	2.21	2.05	2.36	2.39
	(P	0.621	0.740	0.543	1.010	1.04
	(K	0.260	0.32	0.378	0.340	....
% Moist Weight	(N	0.3960	0.456	0.336	0.3565	0.4035
	(P	0.1132	0.1530	0.089	0.1524	0.1835
	(K	0.0478	0.0661	0.062	0.0514	.....

Table 2

Analysis of Hot Extract

Sample	V A	VI A	VII A	VIII A	VIII C	IX C	IX A	IX B
Wt. of material extracted	263.5	260.0	251.	250.	257.5	250.	250.	250.
Dry equivalent	48.5	53.7	41.2	37.9	38.8	44.	44.	44.
Volume Extract cc.	2054.0	2065.0	2070.0	2079.0	2119.0	2095.0	2078.0	2141.0
Solids								
Mgms per 100cc.	480.	614.	565.	587.5	626.	621.	649.	627.
Total gms extracted	12.8	12.7	11.7	12.2	13.3	13.05	13.7	13.3
Percent of dry equivalent	26.4	23.7	28.4	32.4	34.3	29.7	31.1	30.3
Nitrogen								
Total Mgms in manure	1020.	1185.	843.	891.	915.	1050.	1050.	1050.
Mgms per 100cc.	32.6	30.8	27.3	31.5	31.4	34.5	35.6	35.8
Mgms total in extract	669.	636.	565.	655.	664.	725.	752.	757.
Percent in extract	65.6	53.7	67.	73.4	72.6	69.	71.6	72.
Phosphorus								
Total Mgms in manure	306.	398.	223.	382.	392.	457.5	457.5	457.5
Mgms per 100cc.	10.43	12.7	7.38	16.5	15.6	7.55	17.8	17.85
Mgms total in extract	215.	262.	153.	322.	330.	370.	375.	375.
Percent in extract	70.4	65.7	68.6	84.5	84.2	80.9	82.	82.4
Potassium								
Total Mgms. in manure	124.	172.	156.	129.	132.			
Mgms. per 100cc.	15.04	6.94	7.04	5.84	5.9			
Mgms. total in extract	1035.	143.	145.5	123.2	125.			
Percent in extract	83.6	83.3	93.26	95.7	94.56			

Table 3

Analysis of Cold Extract I

Sample	V E	VI E	VII E	VIII E	IX E	IX F	IX G	IX H
Wt. of material extracted	259.0	253.5	272.5	255.0	260.0	260.0	270.0	270.0
Dry equivalent	47.6	52.3	44.7	38.5	45.8	45.8	47.5	47.5
Volume Extract	2027.0	2070.0	2060.0	2100.0	2095.0	2100.0	2078.0	2141.0
Solids								
Mgms per 100cc	480.0	532.0	515.0	524.0	597.0	606.0	641.0	580.0
Total gms extracted	9.75	11.0	10.6	11.0	12.5	12.7	13.3	12.4
Percent of dry equivalent	22.5	21.05	23.7	28.6	27.3	27.7	28.0	24.3
Nitrogen								
Mgms total in manure	1000.0	1155.0	915.0	909.0	1095.0	1095.0	1135.0	1135.0
Mgms per 100cc	22.9	26.7	26.0	27.1	34.0	33.2	36.0	33.7
Total gms in extract	465.0	552.0	536.0	570.0	712.0	697.0	747.0	720.0
Percent in extract	46.5	48.6	58.7	63.0	65.0	63.7	65.7	63.5
Phosphorus								
Total Mgms in manure	296.0	387.0	242.0	389.0	477.0	477.0	49.4	49.4
Mgms per 100cc	8.42	11.4	6.85	15.45	17.20	16.8	21.2	21.1
Total mgm in extract	171.0	236.0	141.0	325.0	361.0	353.0	440.0	452.0
Percent in extract	57.8	61.0	58.3	83.7	75.7	74.0	89.2	91.5
Potassium								
Total Mgms in manure	124.0	167.0	169.0	127.0				
Mgm per 100cc	3.44	6.16	6.55	4.67				
Total mgms in extract	69.8	128.0	135.0	98.0				
Percent in extract	56.3	76.8	80.0	77.2				

Table 4

Analysis of Cold Extract II

Sample	V E	VI E	VII E	VIII E
Wt. of material Extracted, Gms.	259.0	253.5	272.5	255.0
Dry equivalent	47.6	52.3	44.7	38.5
Vol. extract cc.	2040.	2070.	2040.	2105.
Solids				
Mgms. per 100cc.	216.5	255.	174.	160.
Total Gms.extract	4.42	5.3	3.6	3.36
% dry equivalent	9.21	10.11	8.05	8.74
Nitrogen				
Mgms. total in manure	1000.	1155.	915.	909.
Mgms. per 100cc.	11.	13.6	7.08	6.93
Mgms. total in extract	224.	282.	144.	146.
% in extract	22.4	24.4	15.75	16.10
Phosphorus				
Mgms. total in manure	296.	387.	242.	389.
Mgms. per 100cc.	1.55	1.6	3.63	2.23
Mgms. total in extract	30.6	33.	74.0	44.7
% in extract	10.3	8.53	30.6	11.5
Potassium				
Mgms. in total manure	124.	167.	169.	127.
Mgms. per 100cc.	2.30	1.51	1.47	1.72
Mgms. total in extract	43.4	31.3	30.1	51.5
% in extract	35.1	18.7	17.81	20.3

Table 5

Analysis of Residue after Hot Extraction

Sample		VA	VIA	VIIA	VIIIA	VIIIB	IXC	IXA
Total dry equivalent extracted (gm.)		48.5	53.7	41.2	37.8	38.8	44.0	44.0
Total solid in extract (gm.)		12.8	12.7	11.7	12.2	13.3	13.05	13.7
Residue by difference (gm.)		35.7	41.0	29.5	25.6	25.5	30.95	30.3
Residue by weight		....	....	30.5	25.2	26.0	28.2	28.9
Percent of Residue	(N	1.07	1.29	0.965	0.956	0.950	1.28	1.210
	(P	0.229	0.347	0.248	0.251	0.253	0.303	0.291
	(K	0.057	0.0952	0.037	0.0217	0.0281	.....	.....
Total in Manure	(N	1020.	1185.	843.	891.	915.	1050.	1050.
	(P	306.	398.	223.	382.	392.	457.	457.
	(K	124.	172.	156.	129.	132.	....	....
Total in Residue	(N	382.	529.	284.	245.	252.	398.	367.
	(P	81.6	138.	73.2	64.2	65.5	94.	87.1
	(K	20.3	39.	10.5	5.55	7.17	....	.....
Percent in Residue	(N	34.5	44.6	33.7	27.5	27.5	37.9	34.9
	(P	26.7	34.7	32.8	16.8	16.7	20.5	19.1
	(K	16.4	16.8	6.74	4.3	5.44	....	....

Table 6

Analysis of Residue after Cold Extraction

Sample		V E	VI E	VII E	VIII E	IX F	IX G
Total Solids Ext.I(gms.)		9.75	11.00	10.6	11.0	12.7	13.3
" " " II "		4.42	5.3	3.6	3.36	....	....
" " in Extracts		14.17	16.3	14.2	14.36	12.7	13.3
Total dry equivalent extracted (gms.)		47.6	52.3	44.7	38.5	45.8	47.5
Total Solid in Extracts (gms.)		14.17	16.3	14.2	14.36	12.7	13.3
Residue by difference		33.43	36.6	30.5	24.14	33.1	34.2
" " weight		.....	.....	26.3	23.5	29.4	34.2
Percent of Residue	Nitrogen	1.04	0.996	0.855	0.8850	1.405	1.37
	Phosphorus	0.301	0.496	0.107	0.0950	.389	.210
	Potassium	0.031	0.0205	0.0121	0.0136	.....	.....
Total in Manure	Nitrogen	1000.	1155.	915.	909.	1095.	1135.
	Phosphorus	296.	387.	242.	389.	477.	494.
	Potassium	124.	167.	169.	131.	....	....
Total in Residue	Nitrogen	348.	364.5	261.	213.5	485.0	468
	Phosphorus	100.	128.0	32.6	22.9	128.7	71.8
	Potassium	10.3	7.5	3.7	3.28	.....	.....
Percent in Residue	Nitrogen	34.8	31.5	28.5	23.5	42.4	41.2
	Phosphorus	33.8	33.1	13.5	5.9	27.0	14.5
	Potassium	8.3	4.5	2.19	2.5	....	....

The short summary table below indicates the comparative results of the study of the methods of preparation much better than an extensive discussion.

Table

Percent of Fertilizing Constituents by the various methods of extraction

Hot Extraction

Sample		VA	VIA	VIIA	VIIIA	VIIIC	IXA	IXB	IXC
%	N	65.5	53.7	67.0	73.4	72.6	71.6	72.0	69.0
	P	70.4	65.7	68.6	84.4	84.2	82.0	82.4	80.9
	K	83.6	83.2	93.2	95.7	94.5	....	....	....

First Cold Extraction

Sample		VE	VIE	VIIE	VIIIE	IXE	IXF	IXG	IXH
%	N	46.5	48.7	58.7	63.0	65.0	63.7	65.7	63.5
	P	57.8	61.0	58.3	83.7	75.7	74.0	89.2	91.5
	K	56.3	76.8	80.0	77.2	....	....	....	....

Second Cold Extraction after fermentation

Sample		VE'	VIE'	VIIE'	VIIIE'
%	N	22.4	24.40	15.75	16.1
	P	10.3	8.53	30.60	11.5
	K	35.1	18.70	17.81	20.3

Total of First & Second Cold Extractions

Sample		V	VI	VII	VIII
%	N	68.9	73.10	74.45	79.1
	P	68.1	69.53	88.90	95.2
	K	91.4	95.50	97.81	97.5

Only the most general conclusions regarding the various methods of extraction can be drawn. The hot extraction usually removes from 5% - 10% more of the constituents, nitrogen and phosphorus, and from 10% - 15% more of the potassium than does the immediate cold extraction. However the sum of the percentages obtained by the two cold extractions is somewhat greater than by the single hot extraction.

Very little difference is to be noted as a result of the different methods of making the hot extract.

Cold extraction after fermentation removes a slightly greater percentage of phosphorus than an immediate cold extraction.

Fermentation Changes

Having determined the relative proportions of total nitrogen, phosphorus and potassium in liquid manure as affected by different methods of preparation, it was desirable to investigate the bio-chemical changes which take place with respect to nitrogen in fermentation of the extract since these changes may affect the availability of the nitrogen and factor in the utility of this method of preparation.

Accordingly, 870 grams of fresh cow manure was treated with eight liters of water, thoroughly shaken and an extract of 7,225cc. filtered off.

Ammonification in Solution

A portion of this extract was set aside and allowed to ferment in an eight liter stoppered bottle at room temperature. At the intervals stated in the table below the free ammonia was determined by aeration of 100cc. portions with 4 grams sodium carbonate. The results obtained are indicated below:

Table VII

100cc. original extract = 35 mgms. N as NH_3.

Days Fermentation		0	4	7	11	14
Sample						
Mgms. NH_3 per 100cc.	a	4.76	5.4	9.10	7.82	7.38
	b	4.79	5.5	9.35	7.88	7.40
Mean		4.77	5.45	9.17	7.85	7.40
% Ammonification of total N		13.61	15.55	26.20	22.40	21.15

Ammonification of the solution reached a maximum on the seventh day. The decrease in the amount of ammonia after the seventh day is probably due to volatilization.

Ammonification of the Fresh Extract in the Soil

The question of the rate of ammonification of the extract in the soil is of great importance from the feeding standpoint.

The beaker method of study of ammonification has been adapted by Lipman[3] and others as a means of determining the availability of organic fertilizers, and was employed here.

Twenty 100gm. portions of the soil, which had been taken from section No. 163 (see page 51) and allowed to air dry in the laboratory, were weighed into jelly glasses. To eighteen of these 25cc. of the manure solution was added. They were weighed, then covered with watch glasses and allowed to incubate at room temperature. Moisture content was maintained by making up to weight with distilled water at the end of each week. Every three or four days duplicate samples were taken, and on a 50gm. portion ammonia was determined by the aeration method of Potter and Snyder,[4] using 5 gms. of magnesium oxide as a base instead of sodium carbonate, and doubling the quantities of soil and water which they recommend.

The results given in table VIII show that the ammonification was at a maximum within seven days. The rate of transformation of the organic nitrogen is thus shown to be rapid and the proportion changed is very large.

Although the study of the aeration method (page 39) indicates that the percent of ammonia obtained varies with the quantity of soil used, still if the quantity withheld by the same quantity of soil is constant under the same conditions of the determination, the subtraction of a blank soil as indicated in the table should give the quantity of ammonia produced with some accuracy. Potter and Snyder report practically complete recovery of small additions of ammonium sulfate to soil by the aeration method.

Table VIII

Ammonification and Nitrification of Fresh Extract in Soil Culture

25cc. Extract = 8.75 mgms. Nitrogen as NH_3.
25cc. Extract applied to 100 gms. soil.

Days incubation	Sample	Mgms. Ammonia per 50gms.	Mgms. Ammonia mean per 100gms.	Mgms. Ammonia minus check	Mgms. Ammonia %total N as NH_3	Mgms. Nitrate Expressed as NH_3 per 50gms.	Mgms. Nitrate Expressed as NH_3 mean per 100gms.	Mgms. Nitrate Expressed as NH_3 minus check	Mgms. Nitrate Expressed as NH_3 %total N as NH_3
4	A. 2.38 B. 2.22	4.60	2.85	32.6	2.07 2.19	4.26	0.33	3.85
7	A. 4.78 B. 4.87	9.65	7.9	90.3	2.61 2.37	4.98	1.11	12.7
11	A. 2.66 B. 2.55	5.21	3.46	39.6	3.08 2.75	5.83	1.96	22.1
14	A. 2.42 B. 2.44	4.86	3.11	35.6	2.55 2.89	5.44	1.61	18.4
18	A. 1.49 B. lost	2.98	1.23	14.0	2.5 3.3	5.8	1.97	22.5
21	A. 1.26 B. 1.25	2.51	0.76	8.68	5.08 5.27	10.35	6.52	74.5
25	A. 1.13 B. 1.15	2.28	0.52	6.06	....	.....	....	....
29	A. 1.09 B. 1.13	2.22	0.47	5.37	....	.....	....	....
32	A. 0.95 B. 0.99	1.94	0.19	2.17	4.78 4.44	9.22	5.39	61.6
Check on Soil. 0.	A. 0.91 B. 0.84	1.75	....	....	1.92 1.95	3.87	....	....

Nitrification

Nitrates were determined by the following modification of the Devarda-Allen method[5] as adapted for use in the soil biology laboratory of this University.

The soil was dried at 100-108 degrees for ten hours, then placed in a shaker bottle, 300cc. of dilute hydrochloric acid, (5cc. sp.gr. 1.20 to one liter) added, and the bottle shaken for several minutes.* After allowing the soil to settle, a 200cc. portion of the solution was drawn off, placed in a Kjeldahl flask and 8cc. of a solution of potassium hydroxide added. (The alkali solution was made by dissolving 300gms. of potassium hydroxide in 1200cc. of distilled water, 10-15 strips (5-8 gms.)of aluminum were then added and after they had dissolved the solution was boiled down to a liter).

After adding the alkali to the soil extracts the free ammonia was boiled off, the volume made up to 200cc. with ammonia, free water, and about one half gram of Devarda alloy added. The extracts were then distilled for about an hour and the ammonia produced in the reduction, received in standard acid.**

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* The methods of extracting nitrates havebeen given considerable study by Lipman and Sharpe[6] and they have adopted calcium oxide as a defloculating agent. A comparative study of calcium oxide with calcium carbonate by Potter and Snyder[7] seems to throw doubt upon the advisability of the use of calcium oxide but good results are reported with the use of calcium carbonate. In the soil biological laboratory here, dilute hydrochloric acid is considered to give the most complete recovery of nitrates and the extract may be drawn off after a brief settling without filtering.

** The more elaborate apparatus of Allen would undoubtedly give more accurate results than the straight distillation as carried out here. However if the boiling is maintained very gently and the more rapid distillation begun, only when the reduc-

The results of the nitrate determinations are also given in table VIII. The production of nitrates reached a maximum on the 21st day. As can be noted in the table, the sum of the nitrates plus ammonia on the seventh day equals 103% respecting the total nitrogen added. However a small amount of the nitrogen originally in the soil may have been transformed to nitrate.

Some variation in the nitrates of duplicate soil samples is to be expected since nitrate formation is dependent on type of organism and variation in the factors governing its activity, such as aeration and moisture, may vary considerably even in duplicate samples of the same treatment.

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tion is almost complete the carrying over of the alkali is minimized. Furthermore if precaution is observed in the boiling the end point of the titration of the evolved ammonia, which usually gives trouble is much more satisfactory, in fact in most cases is quite distinct.

Ammonification of Fermented Extract in Soil

Since, after a couple of weeks of fermentation of the extract solution, the ammonification under the conditions seemed to have reached a maximum, it was then desirable to see ifammonification would not proceed further in the soil. With this object in view, the soil study was repeated as with the fresh solution and the results were as reported in table IX. They are quite similar to those found for the ammonification of the fresh extract except that the maximum is reached by the third day. The low results for nitrate on the 17th day may be due to over-heating in drying the soil before extraction.

Since the nitrate of the check soil was determined after 21 days a small amount of nitrate formed in the check during incubation gives the small minus results indicated.

On the whole it makes little difference whether the extract be applied fresh or after fermentation. In either case the rate of transformation of the organic nitrogen to ammonia is very rapid and a very large percent of the nitrogen is transformed. A somewhat smaller percent of the nitrogen is eventually changed to nitrate.

Ammonification of Liquid Manure Solution

The ammonification results reported in table VII were made on the filtered extract. The question then arose as to whether the results would have been the same if the extract had not been filtered off but allowed to stand in contact with the residue.

Table IX

Ammonification and Nitrification of the Extract (after two week fermentation) in Soil Culture.

Days incubation	Sample	Mgms. Ammonia per 50gms.	mean per 100gms.	minus check	%total N as NH_3	Mgms. Nitrate N Expressed as NH_3 mean per 100gms.	minus check	%total N as NH_3
3	A. 4.67 B. 4.65		9.32	7.10	81.2	5.10	-0.65	....
7	A. 2.71 B. 2.59		5.20	3.18	36.4	5.47	-0.28	....
10	A. 1.43 B. 1.41		2.84	0.72	8.12	5.60	-0.15	....
14	A. 1.27 B. 1.27		2.54	0.42	4.8	7.84	2.09	23.9
17	A. 1.12 B. 1.07		2.22	0.10	1.14	5.2	-0.55	....
21	A. 1.05 B. 1.07		2.12	0.00	.0	11.9	6.14	70.3
21 Check	A. 1.03 B. 1.09		2.12	....	....	5.75	....	....

Inorder to investigate this point, 870 gms. of fresh cow manure were weighed into each of three 3 gallon bottles, A, B, and C, and made up to a total weight of 8000 gms. by the addition of water. All were shaken thoroughly. The solutions A and C were then filtered through cotton gauze, and the extract returned to the bottle. A was left unstoppered while the other two bottles were stoppered tightly. Ammonia was determined at the end of each week by aeration of 100cc. with 4 gms. of sodium carbonate.

The results given in the table X show but little variation in the quantity of ammonia produced, and there seemed to be no loss from A.

Ammonification of Extracts after
Addition of Acid Phosphates, etc.

In the feeding experiments reported later, acid phosphate was added to the natural extract before applying. The effect of such an addition on the rate of ammonification seemed worthy of study.

There is a current belief among florists that sufficient old manure should be left before the addition of a fresh lot for extraction, in order to secure a more rapid decomposition, and it was desirable to determine if this supposition was justifiable.

It was also interesting to note the effect of a brief heating on the rate of ammonification.

Table X

Ammonification of Liquid Manure

Mgms. Total Nitrogen in 100cc. of Extract as NH_3	A	B	C
	35.9	36.3	35.7

Extract	A			B			C		
Days fermentation	Mgms	NH_3	% Ammonification	Mgms	NH_3	% Ammonification	Mgms	NH_3	% Ammonification
1 (a)	3.11			2.86			2.68		
		3.07	8.55		2.86	7.88		2.65	7.32
1 (b)	3.04			2.86			2.61		
7 (a)	7.80			8.16			6.90		
		7.52	20.92		8.14	22.40		6.92	19.35
7 (b)	7.24			8.12			6.95		
14 (a)	9.67			9.12			8.74		
		9.56	22.64		8.94	24.60		8.67	24.30
14 (b)	9.45			8.77			8.60		
21 (a)	11.50			10.08			11.10		
		11.70	32.30		10.1	27.80		10.91	30.60
21 (b)	11.90			10.12			10.72		
28 (a)	11.43			10.01			11.80		
		11.50	32.10		10.1	27.80		11.76	32.90
28 (b)	11.62			10.20			11.72		
35 (a)	11.12			10.20			11.80		
		11.12	31.10		10.1	27.80		11.68	32.70
35 (b)	11.13			10.01			11.56		

The following experiment was planned to investigate these points:

About 3,200 gms. of fresh cow manure were extracted with 30 liters of water. After allowing the mixture to settle for several hours, about 25 liters of extract were drawn off.

Into 10 two liter flasks the following quantities of extract were measured and the quantity of phosphate indicated, added. To flasks 3 and 4, 100cc. of a rapidly fermenting manure solution was added.

Flask	1	2	3	4	5	6	7	8	9	10
Extract cc.	1000	1000	2000	2000	2000	2000	2000	2000	2000	2000
Acid Phosphate	....	5gms.	....	10gms	....	2.5	5	10	20	70
Ferm.Manure	....	....	100cc	100cc	....	....	....	....	....	....

Flasks 1 and 2 were weighed, boiled for thirty minutes and weighed again. The loss of weight in the 1st flask was 61 gms., in the 2nd, 55gms. This was made up with water.

Ammonification of the Extract.

Effect of addition of phosphates, heating, etc.

The flasks were stoppered, set aside and allowed to incubate, at room temperature. Ammonia was determined on 100cc. samples of each extract at various intervals by aeration with 5 gms of Na_2CO_3 after theaddition of 5 gms of Potassium Oxalate. The results are given in table XI.

Table XI

Ammonification of Extracts.

Effect of Addition of Phosphates, Heating, etc.

Days Incubation	1		4		8		15		25		40	
Flask		Mean		Mean		Mean		Mean		Mean		Mean
1	(a 3.10)	(3.10	3.85)	(3.90	5.61)	(5.65	7.32)	(7.35	6.80)	(6.82	....	
	(b 3.09)		3.94)		5.69)		7.37)		6.84)		....	
2	(a 3.47)	(3.47	3.94)	(3.90	3.96)	(3.82	4.17)	(4.21	4.75)	(4.73	....	
	(b 3.47)		3.86)		3.68)		4.43)		4.70)		....	
3	(a 3.64)	(3.62	6.83)	(6.71	7.07)	(7.09	8.90)	(8.97	9.25)	(9.20	....	
	(b 3.60)		6.60)		7.12)		9.05)		9.16)		....	
4	(a 3.58)	(3.51	5.60)	(5.64	7.12)	(7.16	8.57)	(8.50	8.67)	(8.69	....	
	(b 3.56)		5.68)		7.20)		8.43)		8.71)			
5	(a 3.23)	)3.21	5.60)	(5.52	6.83)	(6.81	7.81)	(7.81	8.60)	(8.54	9.50)	(9.52
	(b 3.19)		5.44)		6.79)		7.81)		8.48)		9.48)	
6	(a 3.42)	(3.33	5.12)	(5.06	5.08)	(5.06	8.36)	(8.30	8.36)	(8.28	7.97)	(7.89
	(b 3.25)		5.00)		5.04)		8.25)		8.26)		7.80)	
7	(a 3.42)	(3.44	4.79)	(4.77	5.20)	(5.26	7.49)	(7.49	7.50)	(7.50	7.60)	(7.60
	(b 3.46)		4.75)		5.32)		7.49)		7.50)		7.60)	
8	(a 3.44)	(3.39	4.83)	(4.77	5.04)	(5.04	6.68)	(6.72	6.66)	(6.64	7.80)	(7.75
	(b 3.34)		4.71)		5.04)		6.76)		6.62)		7.70)	
9	(a 3.47)	(3.45	4.58)	(4.50	4.73)	(4.72	5.82)	(5.78	5.22)	(5.08	4.68)	(4.64
	(b 3.42)		4.43)		4.72)		5.74)		5.04)		4.60)	
10	(a 3.48)	(3.46	4.22)	(4.24	4.22)	(4.22	4.15)	(4.09	3.51)	(3.49	4.22)	(4.16
	(b 3.44)		4.26)		)		4.03)		3.47)		4.10)	

100cc. extract = 27.84 mgms Total Nitrogen as NH_3

1. As can be noted there was a slight loss of ammonia from flask 1 compared with 2, by the boiling.

2. Addition of the fermenting solution to the fresh solution appeared to increase the rate of ammonification slightly.

3. Increasing amounts of acid phosphates depress the rate of ammonification and in amounts exceeding 5 gms per liter almost inhibit action.

Ammonification of the Extracts in the Soil

Forty 100gm. portions of the air dry soil used in previous experiments were weighed out, and to each of four samples, 25cc. of the fresh extract from each flask was added. These were incubated as in previous experiments.

Ammonia was determined on a half portion of two of the soil samples of each treatment at the end of the 4th day. The residue was again set away and allowed to incubate. On the eighth day the ammonia was determined on the other half portion. About 3cc. of water was added to each of the remaining samples at this time. On the thirteenth day these were divided into equal portions and one used for ammonia and the other for nitrate determinations.

The results are given in table XII. The phosphate seems to have caused a slight increase in ammonification in the soil at the end of the fourth day. Very little variation is to be noted in the production of nitrates or ammonia as effected by the different methods of treatment. Unfortunately since the

Table XII

Ammonification and Nitrification of Extracts in Soil Cultures.
Effect of Addition of Phosphates, Heating, etc.

25cc. extract = 6.96 mgs. Nitrogen (as NH_3) per 100gms. soil.

Days Incubation	4		8		13		13	
	Mgms. Ammonia per 100 gms. soil						Mgms. Nitrate per 100gms. soil as NH_3.	
		Mean		Mean		Mean		Mean
Flask 1 (a)	4.58		3.53		3.40			
1		4.48		3.61		3.38		(lost)
1 (b)	4.37		3.69		3.36			
2 (a)	4.95		3.89		3.69		15.0	
2		4.81		3.91		3.65		15.25
2 (b)	4.67		3.93		3.61		15.5	
3 (a)	4.82		4.19		3.16		14.4	
3		4.74		4.02		3.09		13.86
3 (b)	4.66		3.85		3.03		14.32	
4 (a)	4.70		5.01		3.16		13.40	
4		4.78		5.05		3.30		14.07
4 (b)	4.87		5.08		3.44		14.77	
5 (a)	4.46		4.87		3.01		15.92	
5		4.38		4.48		2.98		16.06
5 (b)	4.30		4.10		2.95		16.21	
6 (a)	4.22		3.95		3.16		16.05	
6		4.26		3.98		3.36		16.25
6 (b)	4.30		4.10		3.56		16.95	
7 (a)	4.67		3.64		3.36		16.65	
7		4.71		3.95		3.35		16.37
7 (b)	4.75		4.27		3.33		16.20	
8 (a)	4.87		4.30		3.40		16.22	
8		4.77		4.24		3.51		16.22
8 (b)	4.67		4.18		3.61		lost	
9 (a)	4.82		4.67		4.00		15.85	
9		4.98		4.55		3.72		15.1
9 (b)	5.14		4.43		3.63		15.35	
10 (a)	4.67		4.92		3.48		15.85	
10		4.67		4.94		3.62		15.57
10 (b)	4.67		4.96		3.77		15.30	

experiment was outlined with the idea of comparative results only no blank checks on the soil were run.

Effect of Acid Phosphate on the Retention of Ammonia

Loss of ammonia has been observed in the storage of liquid fertilizers of the urinary type,[8a] and commercial acid phosphate[9a] is one of the substances which has been recommended for preservation.

In order to determine the loss from a fertilizer of the type under investigation, the following procedure was carried out:

Into each of 3 one-liter Erlenmeyer Flasks, 750cc of the fresh liquid extract prepared in the previous experiment were measured. To flask I, 375 gms of acid phosphate was added. The flasks I and II were loosely stoppered with cotton plugs while III was tightly stoppered with a cork. After six weeks the total nitrogen and ammonia was determined in each solution. They were as follows:

	Total Mgms. Nitrogen as NH_3	Mgms NH_3
I	23.05	4.17
II	17.98	1.07
III	25.65	6.16

With both open flasks considerable loss of Nitrogen occurred but a larger percentage was conserved by the addition of acid phosphate.

A Study of the Changes of Organic Nitrogen in the Fermentation of the Manure Extract Under Solution Condition

In recent years the nature of the organic nitrogen in the soil has been given considerable attention. Jodidi,[8] Robinson,[9] Kelley,[10] and others have shown that in the soil the basic nitrogen is a much small proportion of the total nitrogen than in the vegetable proteins.

With sand culture studies of various nitrogenous substances, including a number of pure proteins, Kelley[11] found that the basic nitrogen groups, representing chiefly diamino acids, was most rapidly and extensively decomposed.

The conditions of fermentation of the extract solution are quite different from those which prevail in the soil. Under soil conditions, the bacterial action is chiefly aerobic while in solution it is more nearly anerobic. Ornithin[12] is known to change to putrein and lysin[13] to cadaverine with the elimination of CO_2 by putrefactive bacterial action. However, in soil culture Jodidi found that cadaverine is quite rapidly decomposed with formation of ammonia.[14]

In ammonification studies of casein, dried blood, soy bean cake meal, cotton seed meal and linseed meal, Kelley found

further[11] that with the exception of casein, ammonia formation was greatly retarded under anerobic conditions. This depression under solution conditions as compared to soil conditions has been noted in the study of ammonification of liquid manure.

Since a basic amino acid such as lysin is changed to cadaverine under anerobic conditions, and on the other hand gives ammonia quite readily under aerobic conditions, the question arises as to the probability of the basic group being less susceptible to yielding ammonia under anerobic conditions.

With the question in mind, the study of changes of the organic nitrogen of the liquid manure was undertaken. In this study the Osborne-Harris Modification[15] of the Hausmann method for the determination of protein substances, used by Jodidi and Kelley, was employed.

The following was the general method of procedure:

500cc. of the liquid extract after fermentation was measured into a liter flask, 50cc. hydrochloric acid (sp.g. 1.2) was added and the extract evaporated to about 250cc. on the water bath. 200cc. of the concentrated hydrochloric acid was then added and the solution refluxed for twenty-four hours.

At the end of this time the solution was made up to a liter with distilled water, 200cc. portions were withdrawn and placed in 500cc. Kjeldahl Flasks. These were evaporated almost to dryness on the water bath. To hasten the evaporation a current of air, washed by sulfuric acid to remove ammonia, was passed through the flasks. When evaporation was almost

complete, the flasks were removed and 100cc. of ammonia free water added. A 10% suspension of magnesium oxide was added and the ammonia determined by aeration. After filtering out the humin nitrogen and washing thoroughly with hot water the filtrate was made slightly acid with hydrochloric acid, evaporated to 100cc. and the basic nitrogen precipitated and washed as directed by Osborne.

The fearful bumping in the digestion of the phosphotungstic precipitate which has always been a source of trouble in the determination was finally discovered to be almost eliminated bythe addition of a sufficient quantity of potassium acid sulfate. A melt was obtained which boiled quietly.

The determinations reported below were made on portions of the manure extract prepared in the previous experiment (page 17) and the results are as follows;

	Mgms. of total Nitrogen Expressed as NH_3		% of total Nitrogen Ammonified		% of total Nitrogen decrease
Period Incubation	1 day	7 days	1 day	7 days	
Ammonia	4.77	9.22	12.81	25.1	
Amide	4.38	2.37	11.75	6.45	5.3
Humin	4.43	4.29			
Basic	4.82	2.91	12.90	7.93	4.97
Non Basic	18.85	17.99	50.60	98.9	1.70

As can be noted there is a very high percentage of humin nitrogen, the formation of which is probably due to the presence of considerable carbohydrate material in the extract.

The amide and basic groups of this extract were decomposed at about the same rate and to about the same extent but there was little change in the non-basic group.

The small amount of nitrogen originally present and the range of experimental error in the determination gave difficulty in securing significant difference in the groups, and for this reason the study was not carried further.

Determination of Ammonia in Soil by the Aeration Method

The aeration method has been investigated and applied to practically all classes of determinations of ammonia. Many modifications have been made in an effort to secure satisfactory and reliable results. In some cases it has seemed very impracticable and has been subject to severe criticism.[16]

The determination of ammonia in soil is at best a very unsatisfactory analytical procedure. All methods of extraction have failed to completely recover a definite added amount of ammonia. The distillation of a soil water mixture with magnesium oxide as the base, has been shown to bring about a decomposition of unstable nitrogenous substances to yield ammonia and gives high results. In an attempt to eliminate many of these difficulties Potter and Snyder have adapted the aeration method to this determination and have reported good results

with its use. However, before adoption of the method for use in the present investigation it was thot advisable togive it a preliminary study.

A New Type of Apparatus for Aeration of Ammonia from Soils

Since, in the method of Potter and Snyder, the soil seemed to have a tendency to settle out in the Kejldahl Flask even though the inlet air tube was adjusted as close to the bottom of the flask as possible, a new type of container was devised with the hope of securing a better stirring of the soil.

The general form of the container is indicated in Fig. 1. The cylinder B. is of 60mm diameter and about 300mm length. The opening at the top was of a size to be fitted by a No. 8 stopper, and the inlet tube at the bottom was about 6mm.

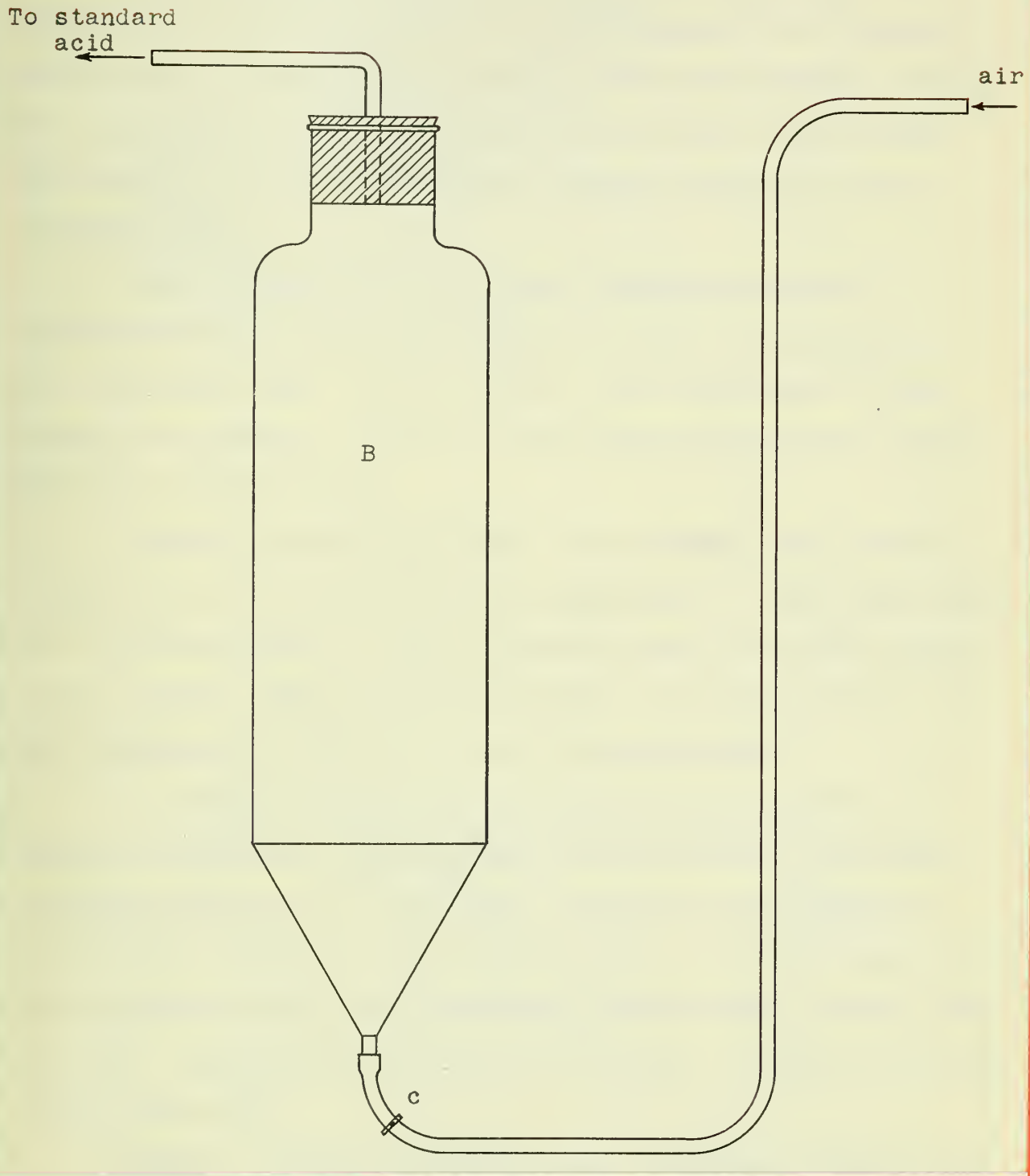
In preparing for the determination, the Hoffman clamp was closed on the rubber tubing at C, water, soil and sodium carbonate added, and the one hole stopper bearing the tube leading to the standard acid was inserted, the clamp was opened slowly and the aeration allowed to proceed in the usual manner.

The advantage of the aeration cylinder over the ordinary Kjeldahl flask in producing a stirring effect is due to the fact that the slope of the sides at the bottom is sufficient to cause the soil particles to slide toward the center where the air is introduced and they are thus kept continually agitated.

Some difficulty was occasioned with the use of the

A New Type of Apparatus
for the
Aeration of Ammonia from Soils.

Fig. I



apparatus as shown in figure I. Some of the larger sand particles would gravitate down into the small air inlet tube and as they accumulated would gradually choke the tube. To avoid this difficulty, the cylinder was sealed at the conical bottom and air was then introduced as into the Kjeldahl flasks. With adjustment of the air inlet tube as close to the bottom as possible the mixing effect was almost as good as in the original design.

The soil used for the ammonia determinations was collected from an unplanted section of one of the benches in the rose house (3a). After drying in the laboratory, it was ground in a mortar, to break up the large particles and passed through a 60 mesh sieve.

Definite quantities of soil were weighed into Kjeldahl flasks as per the method of Potter and Snyder. Distilled water and sodium carbonate in the quantities stated below were added to the flasks, then a drop of kerosene to prevent foaming, and the aerations were carried out in the usual manner.

In the same series, two of the aeration cylinders of special design were connected and the quantities of materials indicated placed in the cylinders. Aeration was allowed to proceed at a rapid rate for about 14 hours. Then the excess of N/50 standard acid in the receiving cylinders was titrated with N/50 KOH, using alizarin red as an indicator. Results are given in the table XIV.

Samples 1-6 were in the Kjeldahl flasks and 7 and 8 were

Determination of Ammonia in Soils by Aeration.
Effect of Varying Quantities of Soil.

Table XIV

Sample	Soil	Distilled Water	Sodium Carbonate	Mgms NH_3
1	25gms	50cc	2gms	.96
2	25	50	2	.87
3	50	100	4	1.12
4	50	100	4	1.12
5	100	200	8	1.34
6	100	200	8	1.25
7	100	200	8	1.24
8	100	200	8	1.26

Table XV

1	25	50	2	1.40
2	25	50	2	1.44
3	50	100	4	2.09
4	50	100	4	2.07
5	100	200	8	lost
6	100	200	8	
7	25	50	2	1.28
8	25	50	2	1.28
9	50	100	4	1.41
10	50	100	4	1.34
11	100	200	8	1.66
12	100	200	8	1.70

in the aeration cylinders.

Even though the inlet air tubes of the Kjeldahl flasks were adjusted as close to the bottom of the flasks as possible, there was not a very good stirring effect on the soil where the larger quantities were used. In the cylinders, the stirring effect was most satisfactory.

Due to the fact that but very small quantities of ammonia were present, a very small difference in the duplicates makes very large percentage error. However, it is very evident that the quantity of soil used affects the relative percent of ammonia yielded in a unit time. It seems probable, too, that the aeration time might have been insufficient. Accordingly the experiment was repeated and this time only the aeration cylinders were used.

Quantities and results are given in Table XV.

Samples 7-12 were disconnected from the series at the end of 24 hours and samples 1-6 were aerated about 20 hours longer. Unfortunately numbers 5 and 6 were lost by foaming over.

The results indicate that small amounts of ammonia are very slowly yielded from the soil by sodium carbonate and both the period of aeration and quantities of soil taken affect the results.

Potter and Snyder state that no appreciable amounts of ammonia are yielded by their method after fifteen hours aeration. They show also that the distillation of the soil with magnesium oxide at 100° gives comparatively large amounts of ammonia on

repeated distillation. Then as a criterion for the stability of easily decomposable nitrogenous compounds respecting loss of ammonia in the aeration procedure, acetamide is taken and subjected to prolonged aeration with sodium carbonate and since only a very small quantity of ammonia is obtained, they conclude that the methodis satisfactory in this respect. However, it has long been known[17] that acetamide does not yield ammonia even on distillation with magnesium oxide at 100° so it would be expected to be stable in presence of sodium carbonate in the cold. However, it is obvious that the smaller quantity of soil (for instance 25 gram portions as used by Potter and Snyder) will give more accurate results. Since the ammonia obtained during an aeration period of 44 hours was greater than that obtained during a period of 24 hours the experiment suggested the use of another base for freeing the ammonia and a comparative study of magnesium oxide and sodium carbonate was consequently made, as follows. See table XVI below. Aeration was in progress for about twenty hours.

Table XVI

Effect of Different Bases

Sample	Soil	Water	Base	Mgms NH_3
1	25	50	2gms $NaCO_3$	1.56
2	25	50	"	1.70
3	25	50	2gms MgO	1.57
4	25	50	"	1.58

From this preliminary experiment it would appear that the choice of base has little to do with the amount of ammonia recovered. Magnesium oxide has this disadvantage: during the aeration the sides of the cylinder become splashed with the oxide-soil mixture and the material does not as readily drain back into the main portion as when sodium carbonate is used.

The experiment was repeated as indicated in the table XVIII. Samples 1,3 and 5 were aerated in cylinders and samples 2, 4 and 6, in Kjeldahl flasks. Sample 1 and 2 were aerated for nineteen hours and 3, 4, 5 and 6 for fifteen hours. The results are given below.

Table XVIII

Effect of different bases and period of aeration

Sample	Soil	Water	Base	Mgms NH_3
1	25gms	50cc	2gms Na_2CO_3	1.09
2	"	"	2gms MgO	1.07
3	"	"	2gms Na_2CO_3	1.11
4	"	"	2gms MgO	1.08
5	"	"	2gms Na_2CO_3	1.07
6	"	"	2gms MgO	1.02

The results check very well. Neither the type of aeration apparatus or kind of base appears to make any difference in the results. Nor does the additional four hours aeration of 1 and 2 in this instance given any significant increase of ammonia.

A Continued Study of the Aeration Method for Determination of Ammonia in Soils. Effect of phosphates.

Since ammonification studies of soils to which large quantities of commercial acid phosphate had been applied were to be made, it was advisable to determine the effect which the phosphate might have on the determination by the aeration method.

The coarsely sieved, air dried soil of the previous experiments was of such low ammonia content that it was decided that an addition of ammonium sulfate solution be made to the soil. The solution contained 6.1 grams of ammonium sulfate to the liter.

It has been found[16] that the presence of magnesium salts and phosphate gives low results in the determination of ammonia by the aeration method, using sodium carbonate as a base. The experiment was then planned to eliminate this difficulty as recommended by Folin[18] by adding potassium oxalate to combine with the alkaline earth metals before adding the sodium carbonate. In addition other bases were tried as outlined in the table XVIII.

Twenty five cc. of ammonium sulfate was added and the phosphate was measured in twenty five portions from a solution and suspension of 10 grams of commercial acid phosphate in 500cc. distilled water, a good shaking and stirring preceeding the withdrawing of each portion. The aeration proceeded over

night. The results of the experiment are given in table XVII, column A.

On the whole the results are about as expected, and the amount of ammonia recovered was practically complete when the strong base was used for the numbers 1-6. The quantity found in 7, 8, and 11 seems abnormally low and may be due to an air leak in the series and consequently incomplete aeration for the last members of the set.

In order to determine just what would be the result of the phosphate acting in solution alone the experiment was repeated and the addition of soil was omitted.

The results given in table XVIII column B show that the quantity of ammonia found in the presence of the phosphate was practically the same for all bases used. It is somewhat surprising that it did not have more influence.

Since the base used did not ap ear to alter the results of the solution tests, it was thought that the low results of numbers 7-12 in the previous experiment (column A of table XVIII) must have been due to incomplete aeration. Accordingly this experiment, in which the soil was added, was repeated with this modification. Two duplicate sets of six members each were operated by separate pumps in order to insure complete aeration. Aeration was in progress for about twenty hours. Unfortunately the second set was lost and only set I is reported in table XVIII, column C. Even though the duplicate set was lost a comparison with column A indicates that the low results of the

Determination of Ammonia in Soils by Aeration

Effect of Different Bases with Phosphate Present

Table XVIII

Series Arrangement	Soil	Ammonium Sulfate Solution	Base	Acid Phosphate Solution	Potassium Oxalate	Mgms NH_3 A	B	C
1	25gms	25cc	2gms MgO	25cc	0	38.2	39.65	37.30
2	"	"	"	"	0	38.2	39.70	.....
3	"	"	2gms CaO	"	0	39.6	39.65	39.00
4	"	"	"	"	0	39.8	39.65	.....
5	"	"	25cc NaOH	"	0	40.30	39.98	42.70
6	"	"	"	"	0	40.10	39.90	.....
7	"	"	2gms Na_2CO_3	"	8gms	34.6	lost	37.80
8	"	"	"	"	"	34.4	"	.....
9	"	"	"	"	0	28.5	39.40	35.75
10	"	"	"	"	0	29.2	39.42	.....
11	"	"	"	0	0	30.3	39.45	37.3
12	"	"	"	0	0	....	39.40	.....

Table XIX

1	25gms	25cc	2gms Na_2CO_3	0	0	39.45
2	"	"	"	0	0	39.25
3	"	"	"	25cc	8	39.25
4	"	"	"	"	8	39.60
5	"	"	"	"	0	38.15
6	"	"	"	"	0	38.05
7	"	"	2gms CaO	"	0	40.60
8	"	"	"	"	0	40.20
9	"	"	2gms MgO	"	0	38.80
10	"	"	"	"	0	38.40
11	"	"	2.5cc NaOH (50%)	"	0	40.80
12	"	"	"	"	0	40.20

latter members in the set A must have been due to incomplete aeration. However, when compared to column B an incomplete recovery is still apparent in most cases. The increased quantities of numbers 5 and 6 column A and C over that of B indicate that the strong base sodium hydroxide must have exercised considerable decomposing action on the organic matter of the soil.

Since the duplicate set of column C was lost it was decided to repeat the experiment. All members were aerated in the same series and the arrangement with reference to the pump was changed to the order indicated in table XIX. A new solution of ammonium sulfate was prepared of about the same concentration used in the previous experiments. Aeration was continued for twenty hours. The results are shown in table XIX in the main check pretty well with previous experiments. On the whole it is safe to conclude that the use of the aeration to determine fairly large quantities of ammonia will give results which will be well within the limit of error in bacteriological studies. Phosphate in the form of commercial acid phosphate decreases slightly the amount of ammonium obtained.

The use of a strong base to liberate the ammonia is, however, contra-indicated, as this strong base would bring about a hydrolysis of more complex forms of nitrogen, with the liberation of undue amounts of ammonia.

Determination of Ammonia in Soil after Ammonification of Casein and Blood Meal in Soil Cultures by the Aeration Method

The previous experiments have shown that the aeration method was quite applicable for the determination of ammonia in soil where fairly large quantities of ammonia were present but that the presence of phosphate gives somewhat low results when magnesium oxide and sodium carbonate are used. However since these bases bring about but little decomposition of organic matter, the aeration method even in view of the fact mentioned above should be applicable to ammonification studies with blood meal and other like nitrogenous substances. The foaming, bumping, and other serious difficulties accompanying the hot distillation with magnesium oxide would be eliminated.

Into each of two liter beakers used four hundred grams of sieved air dried soil was weighed. To beaker 2 four grams of casein was added and to beaker 1, four grams of dried blood meal. These nitrogenous materials were thoroughly mixed with the soil. The beakers were then inoculated with a fresh soil infusion and the moisture content brought up to 30%. After being covered with watch glasses they were set away and allowed to incubate at room temperature. At the end of three and a half days incubation beaker 2 was weighed and showed only a few grams loss of weight by evaporation. After mixing and stirring well, portions were taken out and the ammonia determined by

Determination of Ammonia by Aeration

After Ammonification of Casein.

Effect of An Addition of Acid Phosphate.

Table XX

Sample	Soil	Acid P.	Base	Mgms NH_3
1	50gms	0	5gms MgO	42.5
2	"	0	"	43.0
3	25gms	0.5gm	2.5gms MgO	22.5
4	"	"	"	22.5
5	"	0	"	22.0
6	"	0	"	22.5
7	"	0	"	22.1
8	"	0	"	21.0
9	"	0.5gm	2.5gms Na_2CO_3	19.35
10	"	"	"	19.10
11	"	0	"	21.2
12	"	0	"	21.1
13	"	0	2.5gms MgO	22.70
14	"	0	"	21.00
17	"	0	"	21.60
15	50gms	0	5gms MgO	38.40
16	"	0	"	38.50

Application of Aeration Method to the Determination of Ammonia After Ammonification of Blood Meal in the Soil.

Table XXI

Sample	Soil	Base	Mgms NH_3
1	50gms	5gms MgO	17.20
3	"	"	17.22
5	"	"	16.95
7	"	"	16.85
9	"	"	16.60
11	"	"	16.22
2	25gms	2.5gms MgO	8.97
4	"	"	8.30
6	"	"	8.36
8	"	"	8.30
10	"	"	8.26
12	"	"	8.45

aeration. The quantity of the portions and the methods of treatment are indicated in table XX. Samples 1-12 were aerated in the cylinders for 26½ hours. Samples 13-16 were aerated in Kjeldahl flasks for 21½ hours.

On the whole the results check very well and indicate that the method, tho not absolutely accurate, is quite satisfactory for soil ammonification studies.

At the end of six and a half days the ammonia from the blood meal cultures and the quantities found are shown in table XXI. Aeration was carried out in the cylinders and was continued for 45 hours so that there is no doubt but that all the ammonia possible to be evolved was obtained. As can be noted from the table the results are remarkably uniform considering the chance for variation in the 25 or 50 gram samples. Most gratifying is the checking of the relative amounts of ammonia found regardless of the quantity of soil taken.

Comparative Study of Natural and Commercial Liquid Fertilizers on the Growing of Roses

The following experiment was planned to compare a natural with a commercial liquid fertilizer as affecting the growing of roses.

The University Floricultural greenhouse number 3A was given over to the experiment. The benches in the greenhouse were divided into four foot sections as shown in Fig. II. These sections were filled with fresh soil of the type described

on page 367, Bulletin 176, of the Illinois Agricultural Experiment Station.[19] Well rotted manure was added to the benches at the rate of 1 lb. per square foot (or about 21 tons to the acre), and thoroughly mixed with the soil.

Richmond and Killarney roses were planted in the benches as indicated in the table (a). There were four rows of four plants each in each section. The two rows to the west were of grafted root stock and the two rows to the east were own-root.

Bench II was planted with second year stock, Bench III with third year stock, and Bench IV with first year stock.

Sections of Bench I (261, 262, 263) were not planted but were reserved to obtain soil for bacteriological studies. During the season these sections received watering the same as the planted sections.

A liquid fertilizer equivalent in total content of nitrogen, phosphorous, and potassium, to the average natural liquid manure, can be made by a solution of 300gms. ammonium sulfate, 300 gms. of commercial acid phosphate and 32 gms. of potassium sulfate in 50 gallons of water.

Muncie of this station has found (data as yet unpublished) that the application of acid phosphate gave an increased production in the growing of roses, consequently it was considered advisable to determine the effect on an additional quantity of acid phosphate in the liquid fertilizer.

The following liquid fertilizers were prepared in the following proportions per 50 gallons of water:

Fig. II

Diagram of Green-house 3a

S

E W

N

Bench IV

299	298	297	296	295	294	293	292	291

Bench III

298	288	287	286	285	284	283	282	281

Bench II

279	278	277	276	275	274	273	272	271

Bench I

						263	262	261

Table a

Treatment	Planting Arrangement Bench II	Bench III	Bench IV
Natural Manure	271-R	281-K	291-R
" "	272-K	282-R	292-K
Natural Manure	273-R	283-K	293-R
plus high A. P.	274-K	284-R	294-K
Check	275-R	285-K	295-R
Synthetic Manure	276-K	286-R	296-K
" "	277-R	287-K	297-R
Synthetic Manure	278-K	288-R	298-K
plus high A. P.	279-R	289-K	299-R

R = Richmond
K = Killarney

1. Natural manure.
   20,400 gms. of fresh cown manure.

2. Natural manure plus high A.P.
   20,400 gms. fresh cown manure.
   15,000 gms. commercial A.P.

3. Synthethic manure.
   300 gms. Ammonium Sulfate.
   300 gms. Commercial Acid Phosphate.
   32 gms. Potassium Sulfate.

4. Synthethic manure plus high A.P.
   Same as (3) except for an addition of
   10000 gms. commercial Acid Phosphate.

The solutions were made up in barrels and the various treatments applied as indicated in the table. One application of two gallons of the liquid to the section was made each week, beginning August 29th.

Beginning October 6th, records were taken of the number and quality of the roses produced. The production for the first and second seasons of twelve weeks each is given in the tables, XXII-XXIV.

Discussion of the production of roses and introduction to the study of the biological activity of the soil.

As can be observed from the tables the variation in production is very great and on the whole very inconclusive. However there appears to be a uniform decrease in the production of the sections treated with natural manure plus a high content of acid phosphate below that of the sections treated with natural manure alone. This depression seems most marked during

the first season and particularly in the roses of grafted stock. During the second season the plants became badly attacked by mildew.

The sections at the ends of the house were most seriously affected, which fact may account for the very small number of flowers from sections 271 and 273 during this season.

With the fact of the above decrease in production in mind, it seemed that a study of the bacteriological activity of the soil receiving the various treatments would be interesting and perhaps of value in explaining such differences in production which should prove significant.

Although much biological study of field soils and of pot cultures under greenhouse conditions has been carried on, still so far as is known, no work has been done under greenhouse conditions of floricultural growing. However the same general principles and factors apply.

Since there is no uniform difference between the sections treated with synthetic manure and those treated with synthetic manure plus high acid phosphate, the decreased production noted above could not be due to acid phosphate action alone, but in combination with manure extract, and likely concerned with nitrogen transformation in the soil.

Productions of Roses

Grafted Stock

Table XXII

Treatment	Natural Manure		Natural Manure +High A.P.		Check	Synthetic Manure		Synthetic Manure +High A.P.		Total for bench
Section Variety	271 R	272 K	273 R	274 K	275 R	276 K	277 R	278 K	279 R	
1st Season	87	83	66	74	44	62	65	81	95	657
	170		140			127		176		
2nd Season	45	114	55	109	18	89	42	80	46	598
	159		164			131		126		
3rd Season										

Section Variety	281 K	282 R	283 K	284 R	285 K	286 R	287 K	288 R	289 K	
1st Season	97	106	86	86	63	95	95	95	97	820
	203		172			190		192		
2nd Season	132	78	107	90	85	73	124	83	99	871
	210		197			197		182		
3rd Season										

Section Variety	291 R	292 K	293 R	294 K	295 R	296 K	297 R	298 K	299 R	
1st Season	107	122	98	100	74	96	97	101	111	906
	229		198			193		212		
2nd Season	97	102	99	88	35	107	82	102	84	796
	199		187			189		186		

Production of Roses

Own Root Stock

Table XXIII

Treatment	Natural Manure		Natural Manure +High A.P.		Check	Synthethic Manure		Synthetic Manure +High A.P.		Total for bench
Section	271	272	273	274	275	276	277	278	279	
Variety	R	K	R	K	R	K	R	K	R	
1st Season	71	73	70	73	41	82	53	68	61	592
	144		143			135		129		
2nd Season	16	89	39	99	22	96	35	101	57	554
	105		138			131		158		
Section	281	282	283	284	285	286	287	288	289	
Variety	K	R	K	R	K	R	K	R	K	
1st Season	109	97	89	83	50	74	61	77	107	747
	206		172			135		184		
2nd Season	113	62	98	77	81	60	106	61	104	762
	175		175			166		165		
Section	291	292	293	294	295	296	297	298	299	
Variety	R	K	R	K	R	K	R	K	R	
1st Season	79	86	71	81	57	73	52	73	62	634
	165		152			125		135		
2nd Season	51	73	55	81	22	87	42	89	37	537
	124		136			129		126		

Total Production of Roses

Table XXIV

Grafted

Treatment	Natural Manure	Natural Manure +High A.P.	Check*	Synthetic Manure	Synthetic Manure +High A.P.
First Season	602	510	181	510	580
Second Season	568	548	138	517	494
Third Season					
Total	1170	1058	319	1027	1074
			Own Root		
First Season	515	467	148	395	448
Second Season	404	449	125	426	449
Third Season					
Total	919	916	273	821	897
			Grafted and Own Root		
First Season	1117	977	329	905	1028
Second Season	972	997	263	943	943
Third Season					
Grand total	2089	1974	592	1848	1971

* Since the check represents the production of only one section for comparison to the other columns the number in the table should be multiplied by two.

The Effect of Phosphates on the ammonification of Casein.

The effect of phosphates on ammonification of nitrogenous substances has already received some attention. With dried blood in soil cultures, J. G. Lipman[3b] found an increase in the rate of ammonification with the various phosphates in the respective following order: fused phosphate, acid phosphate, basic slag, ground rock, and check, although the differences were very slight.

Fred and Hart[20] found that the potassium non-acid phosphate and calcium phosphate increases ammonification, the increase being greater in the case of the soluble phosphates, especially in the earlier period of incubation. No work was done with commercial acid phosphate.

Further work at New Jersey by MacClean and Wilson[21] has shown that with pure cultures of ammonifying bacteria, acid phosphates produced a depression on ammonification, but with pure cultures of molds, penicillium and others, an increase was observed. Consequently they attributed their increase in the presence of acid phosphates to the mold flora of the soil.

A preliminary experiment to determine the effect of various phosphates acting in the soil of the type used in the green house, was necessary.

Fresh soil from bench 163 in the house 3a was taken and a moisture content of 21% determined after drying in the air over at 100°.

Samples of the fresh soil (122 grams - 100 gms. air dry) were weighed out into glass tumblers and the various soil treatments were made as indicated in table XXV. After the addition of the materials, they were well mixed with the soil. About ten cc. of water was then added to bring the moisture content to the optimum. After covering with watch glasses, the tumblers were set away and allowed to incubate at room temperatures, for four and a half days.

A slight mold growth appeared in most of the tumblers during the period of incubation. At the end of this period the ammonia was determined by aeration. Five grams of magnesium oxide, 100cc. of water and a 25 gram portion of the soil was taken from each sample. The variations in the treatments are slight except in the case of the monbasic acid phosphates where a decided depression is observed. This may be due to the acidity.

Following this preliminary experiment, a study of the biological activity respecting ammonification and nitrification was made on samples of the soil from the sections receiving the various treatments. In this study the general methods used by Brown[22] and others was employed.

Collection of Samples

The collection of the samples of soil from the green-house was made at a time when the soil had become as dry as is generally allowed under the conditions of rose growing. In most of

Table XXV

Effect of Phosphates on Ammonification of Casein

Sample	Fresh Soil	Casein	Treastment	Mgms NH_3	
1	122 gms.	1gm.	2gms $CaCO_3$	111.0	112.5
2	"	"	"	114.0	
3	"	"	0	96.5	98.75
4	"	"	0	101.0	
5	"	"	2gms $Ca_3(PO_4)_2$	110.0	112.0
6	"	"	"	114.0	
7	"	"	2gms $CaHPO_4$	101.0	98.5
8	"	"	"	96.0	
9	"	"	2gms $CaH_4(PO_4)_2$	49.2	48.4
10	"	"	"	47.6	
11	"	"	2gms Com. Acid P	99.0	97.0
12	"	"	"	95.0	

the sections it was still quite moist. The last application of liquid fertilizer had been made five days before.

The samples were collected in the following manner. The first half inch of soil was scraped to one side on two different areas about six inches square. The soil portions were taken to the entire depth of the bench by means of a sterile cultivating trowel. They were placed in glass jars and taken to the laboratory where they were emptied out on to separate pieces of heavy glazed paper, mixed thoroughly and then returned to the jar.

On 100 gram portions, moisture was determined by drying at 100 degrees. Nitrates were determined on these dried portions. Ammonia was determined on a 50 gram dry weight equivalent of the fresh soil by aeration in the usual manner.

The results given below show the presence of very little ammonia in the soil and a considerable variation in moisture and nitrates.

Section	281	282	283	284	285	286	287	288	289
Moisture %	31.5	25	25	26.5	22.5	25	28	22	28
Mgms Nitrate as NH_3 per 50 gms soil.	4.85	5.14	4.24	3.59	2.16	3.67	4.50	4.97	5.63
Mgms NH_3 per 50 gms. soil	1.23	1.26	1.49	1.23	1.17	1.17	1.48	1.32	1.59

Ammonification Capacity of the Soil

Duplicate portions of the fresh soil from each section, equivalent to 50 grams dry weight, were weighed into jelly glasses. One gram of dried blood was added to each, the samples thoroughly mixed, and the moisture content brought up to 30%. After incubation for six days at room temperature, ammonia was determined on the half portion from each sample. Results are given in the table XXVI.

Sections 283 and 284 show a very much higher ammonifying capacity than the others. This is to be expected since manuring and the addition of acid phosphate tend to increase the rate of ammonification.

Nitrification Capacity of the Soil

Duplicate 100 gram portions of the air dried soil from the different sections were weighed into jelly glasses, 2cc of 5% ammonium sulphate added, and 25cc of the fresh soil infusion from their respective sections was added. The samples were incubated for two weeks and nitrates were determined. Results given in table XXVII seem to indicate a slight decrease in nitrate formation in all sections fed with liquid fertilizers having a high content of acid phosphate.

The depression in production noted, if it really be significant, cannot then be explained on this fact alone. It may be that the presence of the large amount of phosphate along

Table XXVI

Ammonification Capacity of Bench Soils

Section of Soil Sample		Mgms of Ammonia per 25 gms.	Mean of Duplicates	Mean of Sections receiving same treatment	Mgms. Ammonia per gm. Dry Blood
281	(a)	31.0	31.8		
	(b	32.6		28.05	56.10
282	(a)	25.4	24.3		
	(b	23.2			
283	(a)	44.4	40.6	44.87	89.74
	(b	35.8			
284	(a)	50.6	49.15		
	(b	47.7			
285	(a)	30.9	30.65	30.65	61.30
	(b	30.4			
286	(a)	28.4	26.05		
	(b	23.7		26.00	52.00
287	(a)	27.6	25.95		
	(b	24.3			
288	(a)	28.2	26.65		
	(b	25.1		26.50	53.00
289	(a)	27.8	26.35		
	(b	24.4			

Table XXVII

Nitrification Capacity of Soil

Section		Mgms Ammonia Nitrified Per 100 gms	Mean of Duplicates of same section	Mean Of sections receiving the same treatment	Minus Original Nitrate
281	a	8.12	8.39	9.77	4.77
	b	8.65			
282	a	9.87	11.14		
	b	12.40			
283	a	8.98	8.27	7.82	3.90
	b	7.65			
284	a	7.52	7.36		
	b	7.19			
285	a	6.00	5.89	5.89	3.73
	b	5.77			
286	a	10.81	10.67	10.84	6.76
	b	10.52			
287	a	10.80	11.00		
	b	11.20			
288	a	8.45	8.16	8.61	3.31
	b	7.87			
289	a	9.25	9.05		
	b	8.85			

with the organic matter in the natural manure section increased the building up of nitrogen into bacterial protein substances and consequently prohibited some of the nitrogen from becoming available to the plants. It has already been stated that with data of but a single year available and this data showing so great individual variations no definite conclusions can as yet be drawn regarding the production of roses as a result of the different treatments. Nevertheless it is believed that the accompanying study of the biological activity of the soil may furnish light on the results to be obtained.

In the case of farm crops, Brown[23] has noted a correlation between the bacterial activities of the soil and crop yields and he even goes so far as to venture the tentative conclusion that "the tests of such bacterial activities (as ammonification, nitrification and azofication, may indicate quite accurately the crop producing power of a soil or at least the relative crop producing power of several soils." On the other hand Allen and Bonazzi[29] states that the"nitrifying power of a soil may or may not correlate with its crop producing power." Furthermore they conclude that "at present, speculations in regard to relations between soil fertility and nitrification, nitrifying powers, etc. are only speculations." However as concluded by Green[25] the comparison of the results of manuring experiments in the field with those of laboratory tests may be of considerable value in affording information as to the decomposition processes naturally occurring in soils."

III. Summary.

The different methods for the preparation of liquid manure from fresh cow manure do not show uniform differences as to the percentage of total nitrogen, phosphorus, and potassium which they extract. In most cases from 50 percent to 80 percent is extracted and the hot extraction is from 5 percent to 10 per cent more efficient than the cold extraction. A comparatively small amount of the fertilizer constituents is obtained by a second cold extraction.

The extracts contain on an average about 30 mgs of nitrogen, 12 mgs of phosphorus and 7 mgs of potassium per 100cc.

Ammonification of the liquid manure in solution reaches a maximum in about two weeks at the temperature of the laboratory. About 25-30 per cent of the total nitrogen is transformed to ammonia. Commercial acid phosphate in quantities about 10 grams per liter almost inhibited ammonification. In amounts of 5 grams or less per liter there was no serious retardation of the production of ammonia.

In soil cultures, ammonification of the extract reached the maximum in from 4-7 days. A very high percentage of the nitrogen (about 80 per cent)was changed to ammonia. Commercial acid phosphate did not exert much influence on the ammonification of nitrification in the soil culture.

The aeration method for the determination of small amounts of ammonia in soil does not give proportionately increasing

amounts of ammonia with increasing quantities of soil. Although the method can not be considered accurate for small amounts of ammonia, it is probably as nearly so as any devised. In bacteriological ammonification studies where comparatively large quantities of ammonia are determined the aeration method seems very applicable.

The comparative study of the production of roses with natural and commercial liquid fertilizers, as yet shows practically no significant differences in the number of flowers produced.

In the sections treated with natural manure plus a large amount of acid phosphate, the ammonification capacity of the soil was almost double that of the sections receiving the other treatments, and there was but little difference between the other sections.

The nitrification capacity of the soil seemed somewhat depressed where large quantities of acid phosphate had been applied.

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